

WHAT IS CLAIMED IS:

1. A method for treating target cells, tissues or pathogens in a subject comprising administering in sequence:

a) a therapeutically effective amount of a non-covalently bound complex to said subject thereby forming a target-tissue-localized complex;

wherein said non-covalently bound complex comprises a multispecific targeting protein comprising at least one target-binding site and one hapten-binding site, and a hapten-enzyme covalent conjugate;

wherein said at least one target-binding site is capable of binding to at least one complementary binding moiety on the target cells, tissues or pathogens or on a molecule produced by or associated with said target cells, tissues or pathogens; and

wherein said hapten-binding site is non-covalently bound to the hapten-enzyme covalent conjugate;

b) optionally, a clearing agent; and

c) a chemotherapeutic drug or prodrug, capable of being converted to a more active drug by the target-tissue-localized complex.

2. The method of claim 1, wherein said multispecific targeting protein is a multispecific antibody or a multispecific antibody fragment.

3. The method of claim 1, wherein said multispecific targeting protein is multivalent.

4. The method of claim 3, wherein said multivalent, multispecific targeting protein is an anti-CEA x anti-indium-DTPA F(ab')<sub>2</sub> x Fab'.

5. The method of claim 1, wherein said multispecific targeting protein is at least bispecific.

6. The method of claim 1, wherein said complex is injected intravenously, intravesically, intra-arterially, intra-tumorally or intraperitoneally into said subject.

7. The method of claim 1, wherein said complex binds to a cellular tumor-associated antigen.

8. The method of claim 7, wherein the cellular tumor-associated antigen is selected from AFP, EGP-1, EGP-2, CD37, CD74, colon-specific antigen-p (CSAp), carcinoembryonic antigen (CEA), CD19, CD20, CD21, CD22, CD23, CD30, CD37, CD74, CD80, HLA-DR, HCG, Ia, MUC 1, MUC 2, MUC 3, MUC 4, EGFR, HER 2/neu, PAM-4, TAG-72, EGP-1, EGP-2, A3, KS-1, Le(y), S100, PSMA, PSA, tenascin, folate receptor, VEGFR, necrosis antigens, IL-2, T101 and MAGE9.

9. The method of claim 1, wherein said hapten-enzyme conjugate comprises at least one hapten.

10. The method of claim 9, wherein said hapten is selected from the group consisting of HSG, DTPA, indium-DTPA, DOTA, indium-DOTA, yttrium-DOTA, fluorescein or biotin.

11. The method of claim 9, wherein two haptens are linked by a peptide of from 2 – 10 amino acid residues in length.

12. The method of claim 9, wherein two haptens are linked by a peptide of from 2 – 5 amino acid residues in length.

13. The method of claim 9, wherein two haptens are linked by a peptide of 3 amino acid residues in length.

14. The method of claim 9, wherein the haptens are attached via a single reaction site to the enzyme.

15. The method of claim 1, wherein the enzyme is an esterase, amidase, glucuronidase or a galactosidase.

16. The method of claim 15, wherein the enzyme is a carboxylesterase.

17. The method of claim 16, wherein the carboxylesterase is rat, mouse, rabbit, porcine or human carboxylesterase.

18. The method of claim 15, wherein the enzyme is produced by recombinant techniques.

19. The method of claim 18, wherein the enzyme is produced in yeast, bacteria, plants, insect cells or animals.
20. The method of claim 15, wherein the enzyme has been modified to enhance its catalytic properties.
21. The method of claim 20, wherein the enzyme is modified by site-directed mutagenesis.
22. The method of claim 20, wherein the modification increases the rate of enzyme-substrate catalysis and/or reduces the Michaelis constant of the enzyme.
23. The method of claim 1, wherein the multispecific targeting protein binds to both its antigenic target and to its hapten target with an dissociation constant of at least  $10^{-7}$ , more preferably at least  $10^{-9}$ .
24. The method of claim 1, wherein the multispecific targeting protein is murine, chimeric, humanized, human, or a mixture of proteinaceous components from this list.
25. The method of claim 1, wherein the optional clearing agent is an antibody directed against an epitope of the multispecific targeting protein/hapten-enzyme complex.
26. The method of claim 1, wherein the optional clearing agent is an anti-idiotypic antibody to the multispecific targeting protein
27. The method of claim 24, further comprising a carbohydrate-derivatized anti-idiotypic antibody to the multispecific targeting protein.
28. The method of claim 25, further comprising a galactosylated anti-idiotypic antibody to the multispecific targeting protein.
29. The method of claim 1, wherein the chemotherapeutic prodrug has greater aqueous solubility than the active drug produced by the multispecific targeting protein.

30. The method of claim 1, wherein the chemotherapeutic prodrug is a prodrug of a camptothecin, doxorubicin, taxol, actinomycin, maytansine, calicheamicin or epithilone class of drug.

31. The method of claim 30, wherein the chemotherapeutic prodrug is a prodrug of SN-38.

32. The method of claim 30, wherein the chemotherapeutic prodrug is CPT-11.

33. The method of claim 1, wherein said pathogen is a virus, a fungus, a parasite or bacteria.

34. The method of claim 1, where said subject is a mammal.

35. The method of claim 34, wherein said mammal is a human.

36. A kit comprising, in suitable containers:

a) a multispecific targeting protein, comprising at least one target-binding site and a hapten-binding site, pre-mixed with a hapten-enzyme conjugate; and

b) a chemotherapeutic prodrug,

wherein a) and/or b) optionally further comprise a pharmaceutically acceptable carrier.

37. A kit comprising, in separate, suitable containers:

a) a multispecific targeting protein, comprising at least one target-binding site and a hapten-binding site;

b) a hapten-enzyme conjugate; and

c) a chemotherapeutic prodrug;

wherein said multispecific targeting protein, comprising at least one target-binding site and a hapten-binding site and said hapten-enzyme conjugate are mixed immediately prior to use,

wherein a), b) and/or c) optionally further comprise a pharmaceutically acceptable carrier.

38. A method of making a stable non-covalently bound complex that is capable of localizing to a target cell, tissue, or pathogen comprising admixing a multispecific targeting protein comprising at least one target-binding site and a hapten-binding site, and a hapten-enzyme covalent conjugate;

wherein said at least one target-binding site is capable of binding to at least one complementary binding moiety on said target cells, tissues or pathogens or on a molecule produced by or associated with said target cells, tissues or pathogens; and

wherein said hapten-binding site is capable of stably and non-covalently binding said hapten-enzyme conjugate; thereby making a stable non-covalently bound complex.

39. A method of treating a subject, comprising administering a therapeutically effective amount of a non-covalently bound complex, said non-covalently bound complex resulting from the pre-mixing of said multi-specific targeting protein and a hapten-enzyme conjugate, prior to administration to said subject.

40. The method of claim 1, where said subject is a mammal.

41. The method of claim 40, wherein said mammal is a human.